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## Inflammation through the Ages: A Historical Perspective

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### 1.1 Introduction

Inflammation is older than humanity itself and the earliest signs of inflammatory processes can be found on the bones of dinosaurs. Of course, inflammation has always been accompanying humans since they are on Earth as it can be seen on the bones of the first humanoids and of *Homo sapiens*. The first precise diagnoses of inflammatory disorders were made on Egyptian mummies by *Sir Marc Armand Ruffer* (1859–1917). Accompanying the first British Egyptologists, he gave birth to a new science: “paleopathology.” He made a pioneer post-mortem diagnosis of arthritis and spondylitis, and by studying the mummy of Ramses II, he diagnosed that the pharaoh had suffered from atherosclerosis. In fact, long before inflammatory processes could be understood or even defined humans had proposed various therapeutic approaches to treat different types of inflammatory diseases.

### 1.2 The First Treatments

According to Chinese mythology, herbology or the use of plants to cure diseases was introduced by Emperor *Shennong* in 2800 BC, and the first ever book on medicinal plants was published in China in 300–200 BC. Other testimonies of interest on plants to cure diseases or at least to relieve pain and fever were provided by *Edwin Smith* and *Georg Moritz Ebers*, two Egyptologists who obtained fascinating papyruses. These papyruses (around 1520 BC) were copies of even older ones (3400 BC). Not only did they describe case reports of injuries but also listed different plants to be used against various types of injuries (crocodile bites, burns, fractures, bowel diseases, joint pains, etc). For example, infusion of dried myrtle was recommended for rheumatic pain. The interest to use plants to cure inflammatory diseases was perpetuated by the Greeks, and *Hippocrates* (450–370 BC) used extracts from willow bark to relieve pain and

fever. The study of willow bark ended with the discovery of aspirin in the nineteenth century.

Hippocrates also advocated bloodletting as another therapeutic approach to cure most diseases, including inflammatory disorders. Its use was supported by other erudite Greeks such as *Erasistratus*, *Asclepiades of Bithynia*, or *Galen of Pergamon* and later by the Roman scholar *Aulus Cornelius Celsus*, the Persian medical doctor *Avicenna* (tenth century), or the Spanish Jewish doctor *Moïse Maïmonide* (twelfth century). All physicians of the kings of France were great supporters of bloodletting. *Ambroise Paré* (1509–1590), the physician of Charles IX, explained why he bled a young man 27 times in four days: “I liked to mention this event, so that the young surgeon will not be too shy to draw blood when confronted to large inflammation.” *Laurent Joubert* (1529–1583), the physician of Henri III, claimed that it was a way to get rid of the “bad blood” while the best was retained. *Charles Bouvard* (1572–1658) had probably prescribed 47 bloodlettings during the last 10 years of Louis XIII who died of Crohn’s disease at the age of 42 years. They used it even when their patients were still young. For instance, *François Vaultier* (1590–1652) bled the young Louis XIV at the age of 9 years when he had smallpox. *Guy Patin* (1601–1672), the dean of the School of Medicine in Paris for a brief period, declared: “There is no remedy in the world that does so many miracles. I have bled my wife twelve times for a pleurisy, twenty times my son for a continuous fever and myself seven times for a cold.” [1] Even on the American continent, bloodletting was popular. Thus, on December 14, 1799, Georges Washington, probably suffering from pneumonia, died when 3.7 l of blood was drained out of his body in one single day. The first doctor to question the usefulness of bloodletting was *Pierre Charles Alexandre Louis* (1787–1872) who in 1835 considered this approach to have had very limited advantages. But pitted against him were leading doctors such as *François Broussais* (1772–1838) who was alleged to have had spilled more blood than Napoleon on all battlefields! If the lancet was commonly used to remove blood from patients, then the use of leeches was another method. According to an estimate, 35 million leeches were used in France in 1830 alone. This trend went unabated until *François-Vincent Raspail* (1794–1878) questioned the method in 1845 saying: “But why resort to violent and bloody means? Do you wish to calm fever? You will not succeed by bleeding [ . . . ] So leave your lancet there, it has made enough troubles since Hippocrates” [2]. In 1856, in Great Britain, *John Hughes Bennet* (1812–1875) also concluded that there was no proven therapeutic advantage of bloodletting.

The other method used to prevent a severe inflammation and infection after a wound was cauterization supported by doctors such as *Giovanni da Vigo* (1450–1525) in Italy and *Paracelsus* (1493–1541) in Switzerland until Ambroise Paré, comparing the relative advantages of cauterization and the use of anti-septics, concluded that the latter were the best. But the word “antiseptic” was coined only in 1750 by *John Pringle* (1707–1782), a Scottish physician, who studied numerous substances able to prevent putrefaction. In 1854, *Florence Nightingale* (1820–1910) advocated hygiene as a means to prevent infection during the Crimean war in order to limit the mortality of wounded soldiers. Of

course, the main advocate of hygiene was *Ignaz Semmelweis* (1818–1865) who succeeded in 1847 to reduce mortality due to puerperal sepsis.

### 1.3 The Definitions

One of the very firsts to define the parameters of inflammation was *Aulus Cornelius Celsus* (25 BC–50 AD), a Roman encyclopedist to whom we owe the famous statement: “*Notae vero inflammationis sunt quatuor: rubor et tumor cum calore and dolore*” (The signs of inflammation are four: redness, swelling, fever and pain). A fifth element was later added “loss of organ function.” Erroneously attributed to Galen of Pergamum, it could have been proposed by either *Thomas Sydenham* (1624–1689) or *Rudolf Virchow* (1821–1902). Of course, inflammation has for a long time been considered a morbid response of the host to any types of insults. However, *John Hunter* (1728–1793), a Scottish surgeon, appropriately defined inflammation in his book published one year after his death: “*Inflammation in itself is not to be considered as a disease, but as a salutary operation, consequent either to some violence or some disease*” [3]. Despite this appropriate definition, one could still read in 1865 in the French dictionary of medicine that “Inflammation is a complex morbid phenomenon, particularly associated with the function of blood circulation.” In his lecture on inflammation, *Elie Metchnikoff* (1845–1916) stated in 1891 that phagocytes were the participant of the inflammatory process and that inflammation should no longer be seen as only being deleterious [4]. Since Metchnikoff, many mechanisms accompanying inflammation have been further deciphered and mediators have been characterized.

### 1.4 Fever

For a while it was believed that fever was consecutive to some obstructions within the blood vessels leading to an accelerated movement within the free vessels. At the beginning of the eighteenth century, a defender of this concept was the Italian physician *Lorenzo Bellini* (1643–1704). *Herman Boerhaave* (1668–1738), a Dutch physician, also thought that increased heartbeats were the source of the accelerated circulation and fever. In 1744, *François Boissier de Sauvages de Lacroix* (1706–1767), while translating in French the book on “haemastatic” written by *Stephen Hales* (1677–1761), added his personal view on fever, confirming the prevailing concept that inflammation was associated with increased blood flow. Thanks to scientists and doctors such as *John Davy* (1790–1868) in the United Kingdom who made the first sets of temperature measurements in different humans and in different environments (1816–1818), *Antoine Becquerel* (1852–1908) in France who invented the pyrometer to measure human temperature (1835), *Thomas Clifford Allbutt* (1836–1925) who invented the clinical thermometer (1866), and *Reinhold August Wunderlich* (1815–1877) of Germany who made more than one million measurements on more than 25 000 patients (1868), normal temperature and fever could be definitively and precisely defined. *William H. Welch* (1850–1934), the first dean of the

Johns Hopkins School of Medicine, offered a wonderful definition of fever in his 1888 Cartwright lecture:

The real enemy in most fevers is the noxious substance which invades the body, and there is nothing to prevent us from believing that fever is a weapon employed by Nature to combat assaults of this enemy. According to this view, the fever-producing agents light the fire, which consumes them. It is not incompatible with this conception of fever to suppose that the fire may prove injurious also to the patients and may require the controlling hand of the physician [5].

What could be these “noxious substances?” The same year, *Nikolai Fedorovich Gamaleïa* (1859–1949), who worked in Odessa and Moscow, spent some time with Metchnikoff at the Institut Pasteur and reported that an injection of dead bacteria could induce fever in rabbits and sheep. Most fascinatingly, he showed that filtered alcoholic extracts of spleen from pyretic sheep could induce fever within 30 min in rabbits [6]. The following year (1889), *Marc Armand Ruffer*, who joined Elie Metchnikoff and Louis Pasteur shortly after the Institute was set up, published a paper with *Albert Charrin* (1856–1907) who was a professor at the prestigious College de France. They reported that the filtered culture of pyocyanic bacillus (*Pseudomonas aeruginosa*) could induce fever in rabbits, in the absence of alive or dead bacteria [7]. Of note, this report was published three years before the German physician and bacteriologist *Richard Pfeiffer* (1858–1945) coined in 1892 the word “endotoxin” that is nowadays widely used. Most fascinating, the authors wrote, although did not prove, that fever was the consequence of the activation of macrophages. In 1894, *Eugenio Centanni* (1863–1942), an Italian pathologist, recognized the intimate relationship between the pyrogenic and toxic properties of the bacterial poison, which he found to be chemically inseparable. This led him to name his material “pyrotoxina” [8]. In 1890, *Hans Ernst August Buchner* (1850–1902) had shown that this material was also pyrogenic [9].

*Valy Menkin* (1901–1960) was the first to attempt to purify the endogenous mediator that could be responsible of fever. In 1943, he isolated a mediator called “pyrexin” [10]. Unfortunately, further analysis of his work suggested that his factor was contaminated with endotoxin [11]. So, in 1953, *Ivan L. Bennet Jr.* (1922–1990) and *Paul Beeson* (1908–2006) were the first to extract a fever-producing substance from rabbit polymorphonuclear leukocytes [12]. In 1955, *Elisha Atkins* (1921–2005) and *W. Barry Wood Jr.* (1910–1971) isolated a circulating endogenous pyrogen in the blood after the injection of typhoid vaccine [13]. In 1984, Charles Dinarello cloned the human interleukin-1 $\beta$ , known to be the endogenous pyrogen among many other activities [14].

## 1.5 Phagocytosis

In 1882, *Elie Metchnikoff* (1845–1916), when he was in Messina (Sicilia) with his family-in-law, made his key observation. He stuck rose thorns into starfish larvae

and was surprised to see that many phagocytic cells in the hemolymph of the starfish surrounded the “foreign object.” He also observed the process in *Daphnia*, which he infected with yeast. These cells were able to move, ingest, and destroy the yeast cells within the water fleas. He then understood that this process was an important mechanism of host defense against infectious agents. When he met his Austrian friend Karl Claus in Vienna, he was offered to publish his observation in a Viennese journal and Karl Claus coined the word “phagocyte” (1883) [15]. It is worth mentioning that this discovery that finally led to a Nobel Prize (1908) was not published in a high-profile journal (happy times!). Interestingly, he was not the first one to observe the phenomenon. In 1847, *Alexander Ecker* (1816–1887) in Germany had described erythrocytes inside rabbit spleen cells, an observation confirmed in 1870 by *Nathanael Lieberkühn* (1821–1887) who reported that leukocytes could ingest erythrocytes. In 1871–1873, *Giulio Bizzozero* (1846–1901) provided the first drawings of macrophages that had ingested erythrocytes. He stated that reticular cells could ingest infective particles that were carried by the lymphatic liquid. He made this farsighted statement: “. . . this fact is, perhaps, the cause of the stoppage of some infections to the lymphatic glands which are connected to the part covered by the infection through the lymphatic vessels” [16]. In 1875, *Sir William Osler* (1849–1919) reported that alveolar macrophages of coal miners were full of carbon particles. In 1881, *Alexander Ogston* (1844–1929) published cartoons of groups of cocci, mostly free, but sometimes in or on large nonnucleated masses of protoplasm of leukocytes [17]. While Metchnikoff was in Messina trying to understand the aim of phagocytosis, *Robert Koch* (1843–1910) identified anthrax bacilli within white blood cells. Yet, Koch had interpreted his finding to mean invasion of the host by bacterial pathogens. The discovery of phagocytosis by Metchnikoff was not easily accepted, however. He recalls: “. . . The controversy about the phagocytosis could have killed me, or sooner permanently weakened me. Sometimes, (I remember such attacks of Lubarsch in 1889, and those of Pfeiffer in 1894) I was ready to get rid of life” (Oct. 1913). But some other scientists were fully convinced. For instance, *Sir Marc Armand Ruffer* was an ardent advocate of phagocytosis in the UK. In 1892, he wrote in the *British Journal of Medicine* [18]: “Should anyone meet a dead lion and find a lamb inside, he, knowing the habits of the lion would not conclude that the lamb had taken refuge in that. True, after a surfeit of lamb, the lion might die of indigestion but the chance of the lamb ever getting out alive would be very small.”

Other sources of phagocytes were also discovered. *Nicolas Tchistovitch* (1860–1926) described that what were then considered epithelial cells were in fact alveolar macrophages [19]. *Carl Wilhelm Kupffer* (1829–1902) described stellate cells in the liver. He incorrectly believed that these cells were an integral part of the endothelium of the liver’s blood vessels. In 1898, *Tadeusz Browicz* (1847–1928), a Polish pathologist, correctly identified them as macrophages, now known as Kupffer cells. In 1922, *Karl Albert Ludwig Aschoff* (1866–1942) introduced the term “reticulo-endothelial system” that included endothelial cells, fibrocytes, histiocytes, and splenocytes, monocytes and Kupffer cells [20]. However, this concept was of limited help since it mixed cells that undergo pinocytosis and cells that are able to achieve phagocytosis.

In 1903, *Sir Edward Almroth Wright* (1861–1947), a British bacteriologist and immunologist who discovered an effective vaccine for typhoid fever, coined the word “opsonisation” from Greek “*οπισονο*” meaning “I prepare victuals for . . .” He nicely explained: “The body fluids modify bacteria in a manner which renders them a ready prey to phagocytes” [21]. But the observation had been first reported in 1895 by *Joseph Denys* (1857–1932), a professor of bacteriology and anatomy at Louvain University. He stated: “In vaccinated rabbit, leukocytes get from sera their power to engulf and destroy *Streptococcus pyogenes*” [22]. In 1904, *Friedrich Neufeld* (1869–1945), a physician and bacteriologist, director of the Robert Koch Institute in Berlin, described the exactly same phenomenon, only based on much more solid experimental works [23]. But the word, “bacteriotropin,” he coined did not survive the effects of time, and was not used after 1930.

In 2003, *Aimee M. deCathelineau* and *Peter M. Henson* coined the word “efferocytosis” derived from the Latin prefix *effero*, meaning “to take away, to put away, to carry to the grave, or to bury” [24]. However, Metchnikoff already knew the process as he had observed: “[ . . . ] many of phagocytes perish and are taken in by other phagocytes, as can be seen in every case a few days after the onset of the inflammation” [25]. But it was described for the first time by *Giulio Bizzozero* in 1871–1872, studying eye inflammation: “In summary, my observation showed the presence of big cells able to engulf white blood suppurative cells or red blood cells in their contractile protoplams. This represents a way through the pus or blood is absorbed from the anterior chamber” [26]. *Marc Armand Ruffer*, while working with Metchnikoff, described in 1890: “Macrophages are able to swallow microphages (neutrophils) and to destroy and digest them” [27].

## 1.6 Diapedesis

*Galen of Pergamon* had considered that the formation of pus was part of the healing process (“*pus bonum et laudabile*,” good and commendable pus). Pus was supposed to facilitate the removal of the unhealthy mood of the injured body. Galen’s theory prevailed for more than a millennium until *Ugo Borgognoni of Lucca* (1160–1257), a surgeon and the founder of the Faculty of Medicine of Bologna, and his son *Theodorico Borgognoni of Lucca* (1205–1298), clergyman and surgeon, critiqued the doctrine in a four-volume work entitled “*Chirurgia*” (1267). In 1810, *Alexandre François Ollivier* (1790–1844), a physician accompanying Napoleon’s campaign, injected himself with the pus from a wound of a severely injured soldier dying from putrid fever and was the first one to demonstrate its contagiousness [28].

In 1845, two scientists studied pus under the microscope and observed that the cells present in the pus were similar to the cells found in blood. *Alfred Donné* (1801–1878), a French bacteriologist and physician, studied blood from a leukemic patient and thought that it might contain pus cells, although he observed a clear-cut difference between these cells [29]. In contrast, *William Addison* (1802–1881), the physician to the Duchess of Kent, appropriately understood that pus cells were derived from white blood cells (or colorless as they called them): “Colorless blood cells are deposited all over the interior of the vessels;

[ . . . ] and at length pass into the tissue; [ . . . ] These facts are all independent of any orifices, rupture or pores in the vessels, allowing the escape of cells” [30]. Yet, the observation of diapedesis was not made. *Augustus Volney Waller* (1816–1870), a British neurophysiologist, was the first to observe natural leukocyte emigration in 1846, using the frog tongue. Hence, the origin of pus cells was not fully demonstrated [31]. In 1858, *Rudolf Virchow* (1821–1902), a medical doctor at Charité Hospital in Berlin, the father of the cell theory, erroneously believed that pus cells were derived from tissue elements following cell division [32]. But his assistant and two of his students made the final demonstration. In 1863, *Friedrich Daniel von Recklinghausen* (1833–1910), a physician and pathologist who left Virchow for a position of professor at the University of Würzburg before joining the University of Strasbourg, induced an experimental keratitis in frogs with silver nitrate. He characterized the pus cells in humor aqueous during acute inflammation and made the seminal observation of the contractility and mobility of colorless cells [33]. In 1867, *Julius Friedrich Cohnheim* (1839–1884) used a combination of colloidal aniline blue injections and microscopy to prove, what Addison had hypothesized, that white blood cells crossed blood vessels to become pus cells [34]. And in 1875, *Julius Arnold* (1835–1915), using an injection of cinnabar to demonstrate the borders of endothelial cells, came to the conclusion that leukocytes moved across blood vessel walls (diapedesis) by passing between endothelial cells at either points of dense staining, “stigmata,” or circles of stain, “stomata” [35]. Quite surprisingly, 33 years after Addison’s work, *Louis Pasteur* (1822–1895) wrote in 1778: “For us currently, it would be the red blood cells that would be the pus cells from a simple transformation from the first into the second” [36]. This illustrates that great scientists can also make wrong statements!

## 1.7 Chemotactism

Nowadays, it is well understood that diapedesis of circulating cells toward tissues is under the control of chemoattractant signals. Interestingly, the concept came from an observation made with bacteria. In 1884, *Wilhelm Pfeffer* (1845–1920), a botanist at the University of Tübingen, Germany, observed bacteria swimming toward the vicinity of the tip of a capillary tube filled with nutrient sugar that had been dipped into a bacterial culture broth. He coined the word “chemotaxis” [37]. The *in vivo* phenomenon was first reported in 1889 by *Cornelis Adrianus Pekelharing* (1848–1922), a Dutch physician and professor of physiological chemistry and histology at the University of Utrecht [38]. He put cotton wool soaked with anthrax bacilli in the peritoneal cavity of a frog. Retrieving this cotton wool some time later, he showed that it contained significantly more leukocytes than those that had been soaked with neutral liquid. He came to the conclusion that bacteria produce chemoattractant factors. The following year, in 1890, *Georges Gabritchewsky* (1860–1907), a Moscovite researcher who spent some time with Elie Metchnikoff, drew the same conclusion [39]. He inserted under the skin of frogs, rabbits or axolotl, small capillary tubes filled with alive or dead bacteria. Twenty-four hours later, the capillary tubes were full of leukocytes,

whereas this was not the case if the capillaries had been filled with saline. The same year in Belgium, *Jean Massart* (1865–1925), a doctor in sciences and medicine working with *Charles Bordet*, the brother of the famous Jules Bordet who got the Nobel Prize for his discovery of the complement, was the first to demonstrate that the host can make chemoattractant factors [40]. He injected bovine bile subcutaneously in a frog. Then, he sampled the transudation liquid and transferred it in a capillary tube into the abdominal cavity of another frog. Twenty hours later, the capillary tube was full of leukocytes, whereas this was not observed with a capillary tube filled with normal lymph. Thus, he had demonstrated that chemoattractant factors were produced by the frog. In 1891, the same team performed another elegant demonstration [41]. They injected s.c. bacteria (*Micrococcus prodigiosus*, nowadays known as *Serratia marcescens*) in a rabbit. Thirty-five minutes later, they sampled the blood, prepared the serum, placed it in a capillary tube and finally transferred it into the peritoneal cavity of another rabbit. Eight hours later, the capillary tube was full of leukocytes, whereas this was not the case when the tubes were filled with normal serum. For the first time, they had demonstrated that chemoattractant factors were produced in response to infection. In 1938, *Valy Menkin* (1901–1960) purified a substance from inflammatory exudates that induced an increased capillary permeability followed by a migration of polymorphonuclear leukocytes. He called his factor “leukotaxine” [42]. Unfortunately, like the case for pyrexin, despite all his efforts, he could not succeed and further studies revealed no convincing evidence that “leukotaxine” existed as a distinct chemical entity [43]! The discovery of well-identified chemoattractant factors, later called chemokines (the word was coined in 1992), was made in 1987 and 1988 with the description of interleukin-8 and macrophage inflammatory protein (MIP), renamed CXCL8 and CCL3 according to the revised nomenclature of chemokines [44].

## 1.8 Infection and Inflammation

The link between microbes, infection, and inflammation was made during the nineteenth century. But some genius had outstanding intuition. This is the case of *Marcus Terentius Varro* (116–27 BC), a Roman scholar who wrote: “There are certain minute creatures which cannot be seen by the eyes, but which float in the air and enter the body through the mouth and nose and cause serious diseases.” In 1822–1823, *Bernard Gaspard* (1788–1871) and *François Magendie* (1783–1855) showed a link between putrefaction and severe diseases, fever, and death [45,46]. In 1863, *Casimir J. Davaine* (1812–1882) was the first one to convincingly demonstrate the link between bacteria and infectious disease, transmitting anthrax with the blood of a sheep containing the bacteria into rabbits which then died following this injection [47]. The same year, Louis Pasteur established that the first six bacteria identified by *Christian Gottfried Ehrenberg* (1795–1876) in Germany, 30 years earlier, were the ferments of putrefaction [48]. Of note, Ehrenberg had coined the word “bacterium” in 1828. Some infectious diseases were not immediately recognized as such. For example, *Antoine de Jussieu* (1686–1758), a French botanist and physician, claimed that puerperal sepsis



was an inflammation of the uterus. He proposed to use extracts of bark of quassia to reduce fever. Interestingly, this plant was well known to Brazilian Indians who used to relieve pain and fever. *Thomas Denman* (1733–1815) in his book, the first ever to be entirely devoted to puerperal sepsis, mentioned that in addition to an inflammation of the uterus, it could be also an inflammation of the bowel [49]. In 1795, *Alexander Gordon* (1752–1799), a Scottish physician who studied the epidemic puerperal sepsis in Aberdeen, also described inflammation of the abdomen, but he was the very first to claim that it was a contagious disease spread by the hand of the physicians [50]. A similar observation was also made in the United States of America by *Oliver W. Holmes* (1809–1894) in 1843 [51]. Nevertheless, the final demonstration was achieved in 1847 by *Ignaz Semmelweis* (1818–1865), a Hungarian doctor working at the maternity clinic of the Vienna General Hospital. He successfully demonstrated that if medical students cleaned their hands after practicing autopsies before helping women in childbirth, the mortality due to puerperal sepsis was significantly decreased. Yet, doctors were not convinced, and he was asked to leave the maternity clinic. He published his key demonstration 10 years later in Hungarian [52]. However, a short note was published in *The Comptes Rendus* of the French Academy of Sciences on February 21, 1848. Ten years after his demonstration, in Paris, *Paul Dubois* (1795–1871), a professor of obstetrics, claimed that the contamination was due to miasma [53]. Eighteen years after Semmelweis' observation, *Carl Mayrhofer* (1837–1882) was asked by Prof. Carl Braun (1862), the head of the same maternity clinic in Vienna, to demonstrate that airborne organisms were the source of childbed fever. After publishing works in agreement with the ideas of his boss, his following works supported Semmelweis' work, concluding that infection was usually the result of contaminated hands [54]. He was fired! In 1869, *Victor Feltz* (1835–1893) and *Léon Coze* (1819–1896), two Strasbourg physicians, were the first to identify the presence of bacteria in the blood of a lady who died of puerperal sepsis [55]. Ten years later, on March 17, 1879, Feltz republished a similar observation [56]. The following day, on 18th March, Louis Pasteur confirmed the observation [57], but challenged the work of Feltz, arguing that his patient had died of anthrax after deliverance.

## 1.9 Usefulness of Inflammation for Adaptive Immune Response

Gaston Ramon (1886–1963), a veterinary doctor, was hired by the Institut Pasteur to prepare horse immune sera against diphtheria toxin and tetanus toxin. He observed that the antibody titers were quite different from one horse to another. He made a link between these titers and the presence of an inflammatory reaction at the site of injection of the antigen as illustrated by the appearance of an abscess. Then, he purposely created an inflammatory reaction at the site of injection using different substances such as tapioca or lanolin. In 1926, he realized that there was a need to involve an inflammatory reaction at the site of injection of the antigen to enhance immune response, and he had discovered the

concept of adjuvant [58]. Many others used different inflammatory substances, such as lipids. For example, *Jules T. Freund* (1890–1960), an Austria-Hungarian born American immunologist, proposed to create a water oil emulsion to which he added inactivated mycobacterium tuberculosis. In search of the smallest bioactive part that allows the adjuvant properties, the biochemist *Edgar Lederer* (1908–1988) and the immunologist *Louis Chedid* joined forces and discovered the muramyldipeptide [59]. The adjuvants, the so-called “little dirty secrets of the immunologists” led *Charles Janeway* (1943–2004) to redefine the immune response [60]. Since then, what used to be defined as nonspecific immunity was renamed innate immunity. Innate immunity and inflammation are more or less two names for a similar mechanism, namely, the defense of the host against infectious or sterile insults.

## 1.10 Mediators of Inflammation

If nowadays the complement system is known to contribute to inflammation, particularly through the release of anaphylatoxins, its first identification was associated with its capacity to induce bacteriolysis as shown in 1894 by *Richard Pfeiffer*. In 1898, *Jules Bordet* (1870–1961), who received Nobel Prize in 1919, identified that hemolysis was similarly induced by serum components, but the word “complement” was coined by *Paul Ehrlich* (1854–1915), who shared the 1908 Nobel Prize with Metchnikoff. Surprisingly, when the word anaphylatoxin was employed in a paper reporting a model of anaphylatoxic shock, its existence was denied. Indeed, an unanticipated severe reaction had been observed in 1901 when *Charles Richet* (1850–1935, Nobel Prize 1901) and *Paul Portier* (1866–1962) involuntarily killed dogs after the second injection of a hypnotoxin from physalia. One of the main mediators of anaphylatoxic shock is histamine. The molecule was synthesized for the first time in 1907 by *Adolf O.R. Windaus* (1876–1959, Nobel Prize 1928), and *Henry H. Dale* (1875–1968, Nobel Prize 1936) who discovered its activity on smooth muscle and on endothelium. In 1937, *Anne-Marie Staub* (1914–2012), who worked in the laboratory of *Daniel Bovet* (1907–1992, Nobel Prize 1936) at Institut Pasteur, was the first scientist to save an animal from a shock induced by histamine. However, the employed antihistaminic drug was toxic, and the first antihistaminic to be used in humans (*Antergan*) was developed by *Bernard Halpern* (1904–1978).

In 1930, *William S. Tillett* (1892–1974) and *Thomas Francis Jr.* (1900–1969), working at the Rockefeller Institute in New York, identified in the serum of a rabbit injected with *Streptococcus pneumoniae* an interaction activity with a carbohydrate extract of the bacteria (called “fraction C”). The C-reactive protein (CRP) was thus discovered and shown to be present in the inflammatory settings independent of any *S. pneumoniae* infection [61]. CRP was purified by *Colin M. MacLeod* (1909–1972) and *Oswald T. Avery* (1877–1955) in 1941 [62] and crystallized in 1947 by *Maclyn McCarty* (1909–1972) [63]. All these investigators were working at the Rockefeller Institute. The pentameric structure was determined in 1977 and its liver origin was demonstrated the following year.

Nowadays, CRP is one of the most common biomarker to confirm the presence of an inflammatory reaction.

In 1935, *Ulf von Euler* (1905–1983) discovered the prostaglandins. This Swedish physiologist and pharmacologist, who also discovered noradrenalin and substance P, was awarded the Nobel Prize in 1970. His father, *Hans von Euler-Chelpin* (1873–1964) was also awarded the Nobel Prize in 1929 for his contribution to the fermentation of sugar and his mother, *Astrid Cleve* (1875–1968), a botanist, geologist, and chemist at Uppsala University, had been the first woman in Sweden to obtain a doctoral degree of science. For their studies on prostaglandins, related biologically active substances, and the discovery that aspirin prevented the production of prostaglandins, the Swedish *Sune K. Bergström* (1916–2004) and *Bengt I. Samuelsson* (1934–) and the British *Sir John R. Vane* (1927–2004) were awarded the 1982 Nobel Prize.

Glucocorticoids was discovered in 1936–1941 by three scientists who were awarded the Nobel Prize in 1950 for their discoveries related to the hormones of the adrenal cortex, their structure, and biological effects. In the United States, *Philip Showalter Hench* (1896–1965) postulated that a natural mediator, which he called substance X, was involved in the improvement of arthritis when patients developed jaundice. He collaborated with *Edward Calvin Kendall* (1886–1972), who purified a large number of substances from adrenals. Among those, was the compound E (dehydrocorticosterone) that was finally considered as substance X [64]. In Switzerland, *Tadeusz Reichstein* (1897–1996), who was born in Poland, also produced numerous substances from adrenals, of which few were bioactive. In 1937, he reported the production of desoxycorticosterone and its first clinical essays [65].

After the identification of the endogenous pyrogen in 1953 as aforementioned, the next cytokine being described was the interferon. In 1954, two Japanese scientists *Yasuichi Nagano* (1906–1998) and *Yasuhiko Kojima* (1928–) noticed that rabbit skin or testis previously inoculated with UV-inactivated virus exhibited inhibited viral growth when reinfected at the same site with live virus. They hypothesized that this was due to some inhibitory factors. They made two major mistakes. The first one was to publish in a French-speaking journal and the second one was not to create a neologism [66]. This explains why the British *Alick Isaacs* (1921–1967) and the Swiss *Jean Lindenmann* (1924–2015), who published three years later in English and coined the word “interferon,” have been regularly considered as the discoverers of this cytokine.

In 1966, a new cytokine was discovered by Bloom and Bennett who illustrated the capacity of the supernatants of T cells involved in delayed type hypersensitivity to prevent macrophage migration (macrophage migration inhibitory factor, MIF) [67]. Most fascinatingly, this cytokine was rediscovered later as a product of the pituitary gland. Interestingly, MIF brought Stanley Cohen to create in 1974 his neologism “cytokine” [68]. The words lymphokine and monokine were commonly used, but Cohen had shown that fibroblasts infected with viruses could release MIF. Then, it was obvious that these mediators were not only the products of immune cells but also a universal language of the cells.

Endogenous pyrogen, osteoclast activating factor, hemopoietin-1, catabolin, lymphocyte-activating factor (LAF), epidermal cell-derived thymocyte-activating

factor (ETAF) are various mediators identified between 1953 and 1981, which appeared to be the diverse biological properties of one molecule, namely, interleukin-1. It was later categorized into IL-1 $\alpha$  and IL-1 $\beta$ , which are members of a family of 11 molecules, including agonists and antagonists [69]. IL-1 bioactivity was the first to be identified in an inflammatory fluid. In 1982, a French dentist working in Joost Oppenheim's laboratory demonstrated the presence of IL-1 in the gingival fluid of patients with periodontal inflammation [70]. In 1986 in Norway, Waage *et al.* reported for the first time the presence of tumor necrosis factor in the plasma of patients with severe meningococcal sepsis [71]. Later on, all pro- and anti-inflammatory cytokines could be detected in the plasma of patients with severe sepsis or sterile systemic inflammation. These measurements illustrate the cytokine storm observed in many severe inflammatory settings, but those are just the tip of the iceberg [72]. Most fascinating is this statement made by William Osler in 1904: "Except on few occasions, the patient appears to die from the body's response to infection rather than from it."

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